

Germination ecology of the clonal herb *Knautia arvensis*: Regeneration strategy and geographic variation

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Abstract. We investigated germination responses and seed recruitment in the clonal grassland herb *Knautia arvensis* (*Dipsacaceae*) throughout its distributional range in Norway. Four predicted relationships between germination responses and field regeneration behaviour were tested using phytotron experiments and experimental and observational field studies. Seedlings appeared in all experimental microsites in the field, corroborating phytotron predictions that gap- or depth-sensing strategies should be absent in the species. Seasonal timing patterns were predicted from a cold stratification response in the phytotron, but these were not supported in the field. The relationship between dormancy, germinability during storage, and seed carry-over in the field largely conformed to expectations. Seeds from four different geographical regions responded differently to temperature and cold stratification and storage. Dormancy and seed carry-over was higher in seeds from a coastal population, where winters are relatively mild and the probability of repeated freeze-thaw events is high, than in populations from mountain and inland areas, where winters are colder. This is discussed against two alternative hypotheses about the relationship between climate and dormancy in seasonal climates.

Keywords: Competition; *Dipsacaceae*; Field experiment; Germination cueing; Microsite; Phytotron; Regeneration niche; Repeated measures ANOVA; Seedling survival; Seasonal timing.

Nomenclature: Lid & Lid (1994).

Introduction

From a life history perspective, the overall importance of the seed and seedling stages of plants are expected to decrease from annual to perennial species and further for species with the capacity for clonal growth (Harper 1977). Seed regeneration has generally been considered a rare event in clonals, primarily linked to initial establishment at new sites. Eriksson (1989) argues that clonal species differ considerably in their rate of seedling recruitment and that this has consequences for their life histories, genetic diversity, spatial structure, and population dynamics. Seed regeneration strategies of clonal species have, however, been relatively little studied (but see Eriksson & Fröborg 1996; Eriksson 1999; Amiaud et al. 2000).

Seed germination and seedling establishment and growth are vulnerable stages in the life histories of plants. Seedlings are smaller, less competitive, less protected against predators and more vulnerable to adverse environmental conditions, such as freezing or deficiency of water or nutrients, than mature plants. Consequently, seedlings are often restricted in their ecological tolerances, an observation that has been formalized in concepts such as 'safe sites' (Harper 1977) or 'regeneration niches' (Grubb 1977; van der Maarel & Sykes 1993). The probability of seedling survival is not constant, however, and survival may be greatly increased if germination can be avoided at times and places where mortality risk is comparatively high. This may occur if seed germination is sensitive to the environment. Such responses, environmental cueing, have been demonstrated in responses to minute changes in temperature regime (Grime et al. 1981; Rice 1985; Schütz 1997; Cavieres & Arroyo 2000), light intensity (Bell et al. 1995) or spectral quality (Batlla et al. 2000; Mandák & Pyšek 2001), soil nutrients (Karssen & Hilhorst 1992; Bell et al. 1995), moisture (Pérez-Fernández et al. 2000) and cold stratification prior to germination (Murdoch & Ellis 1992; Milberg & Andersson 1998), as well as to interactions between factors. If good and bad times

occur stochastically rather than predictably, however, the effectiveness of environmental cueing breaks down. Under such circumstances 'bet-hedging' strategies (Venable & Brown 1988; Philippi 1993a), that leave a fraction of the seeds dormant through periods of good germination conditions (Vleeshouwers et al. 1995), ensure a carry-over of seeds across years and the build-up of persistent seedbanks that may buffer populations against years of high stochastic mortality.

The aim of the first part of this study is to investigate the relationship between phytotron germination responses and field regeneration behaviour in a clonal grassland herb. The following predicted relationships, based on the theory and results outlined above, are tested in our study species *Knautia arvensis*:

Prediction 1: Requirements for light may function as mechanisms for detecting ephemeral bare ground gaps in grassland vegetation (Rice 1985; Olff et al. 1994; Kotorová & Lepš 1999).

Prediction 2: Requirements for fluctuating temperature may function as a gap- or depth-sensing strategies as the daily thermal amplitude decreases under a leaf canopy and with soil cover (Thompson & Grime 1979).

Prediction 3: If freshly shed seeds are dormant with a cold stratification requirement, this will inhibit germination in the autumn and delay germination of newly shed seeds until the following spring (Grime et al. 1981; Baskin & Baskin 1998).

Prediction 4: Dormancy functions as a 'bet-hedging' strategy and will result in carry-over of seeds across years (Philippi 1993a).

For species with large ecological amplitudes, or with wide geographical or altitudinal distributions, the factors and processes controlling seedling mortality risk may not be constant throughout the range. Such differential selection pressures may result in variation in environmental cueing responses or dormancy levels among populations. An area that has received particular attention is the relationship between the cold stratification requirements of populations and local climate. For example, a number of studies have found that the overall effect of cold stratification on germination, as well as the number of months of cold stratification required, increases with the adversity of the winter conditions (Meyer et al. 1989, 1995; Meyer & Monsen 1991; Cavieres & Arroyo 2001). It is argued that untimely germination during the cold season is more detrimental in harsh climates, and that mountain or northern populations therefore put more effort, in terms of adaptation, into avoiding it (Meyer et al. 1989). Other studies have found indifferent (Thompson 1975; Schütz & Milberg 1997) and opposite (Fowler & Dwight 1964; ter Borg 1987) responses, however, and Vleeshouwers et al. (1995) sug-

gest that germination strategies in seasonal climates should be interpreted as responses to the predictability, rather than the severity, of the adverse season. Cold winter conditions, they argue, are actually very good conditions for seed storage and as long as germination at low temperature is prevented, very little effort, in terms of adaptation, is needed to carry the seeds through to the spring. In winter climates that, from the seeds' point of view, are unpredictable; e.g. if the cold season is repeatedly interrupted by warm spells, different sets of germination strategies may be adaptive. This relates to the theory of 'bet-hedging', which predicts that seed dormancy levels and seedbank build up should increase, and annual germination fractions decrease, with stochasticity rather than with the severity of the local climate (Venable & Brown 1988; Philippi 1993b).

The second aim of this study is to test for differences in environmental cueing responses, dormancy levels and potential for seed carry-over among populations of *K. arvensis*. The two alternative hypotheses outlined above; namely that dormancy increases with the severity (*Hypothesis 1*) or with the unpredictability (*Hypothesis 2*) of winter climate, are evaluated by comparing the phytotron germination responses of five study populations (North, Central, Mountain, East and South) from throughout the species' distributional range in Norway.

Material and Methods

The species

Knautia arvensis (*Dipsacaceae*) is a clonal herb widely distributed in Europe and adjacent parts of Africa and Asia (Hultén & Fries 1986). In Norway, it is common in the southern and central lowlands (Nemoral to Middle boreal vegetation zones), and occasional towards the mountains and in the north (North boreal vegetation zone) (Lid & Lid 1994). *K. arvensis* grows in dry meadows, pastures, dry hills, open woods and roadsides. It has a sympodial stock, a taproot and lateral underground stolons. The flowers are arranged in one to several dense capitulae, each containing 55-100 flowers. Self-pollination is restricted by protandry but will, when it occurs, result in viable seeds (Vange 2002). The fruits are single seeded and have an elaiosome, promoting ant dispersal (Tutin et al. 1976; Lid & Lid 1994), as well as hairs which probably promote dispersal by animals. The fruits are denoted as seeds hereafter.

Climatic severity and predictability of the study sites

There are considerable climatic differences between the five sites (Table 1, Fig. 1) in mean annual tempera-

Table 1. Climatic characteristics of the locations of the five study populations. All climatic characteristics are based on data from the 30-yr period 1960-1990 (www.DNMI.no). T_{\min} = mean diurnal minimum temperature and T_{\max} = mean diurnal maximum temperature. *Number of days $T_{\min} < 0^{\circ}\text{C} < T_{\max}$ = mean number of days during the year when the diurnal temperature range spans 0°C (i.e. at least one transition a day from above 0°C to below 0°C , or *vice versa*, see Fig. 1).

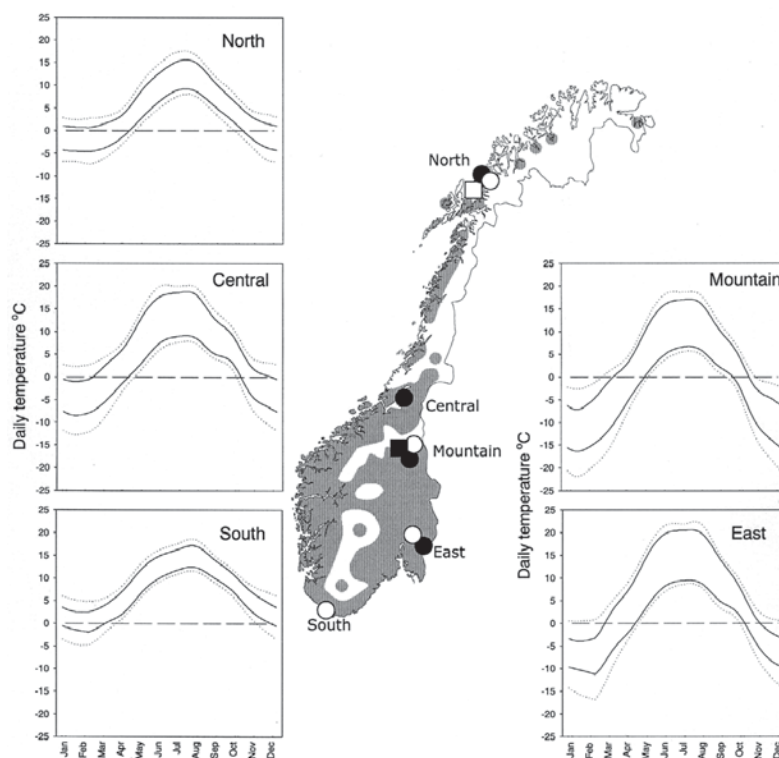
Location	Coordinates	Elevation (m a.s.l.)	Precipitation (mm yr ⁻¹)	Mean temperatures ($^{\circ}\text{C}$)		Number of days $T_{\min} < 0^{\circ}\text{C} < T_{\max}$
				Annual	January	
Mountain	62° 34' N, 11° 05' E	700	504	0.3	-11.2	79
North	68° 50' N, 15° 50' E	5	1032	3.9	-2.5	171
East	60° 01' N, 11° 45' E	200	665	4.0	-6.9	76
Central	63° 47' N, 11° 20' E	50	855	4.6	-4.2	111
South	58° 05' N, 06° 47' E	20	1147	7.4	1.0	80

tures, seasonal patterns and within and between year patterns in the daily minimum and maximum temperatures. *Hypothesis 1* states that dormancy should increase with the severity of winter climate. By using mean January temperatures, which vary considerably between populations (-11.2°C to 1.0°C), as an estimate of winter severity the following rank order in dormancy can be predicted: Mountain > East > Central > North > South. *Hypothesis 2* states that dormancy increases with the unpredictability of the winter climate. Following the argument outlined in the Introduction, we use duration of the period when the diurnal temperature fluctuation spans 0°C (i.e. when there is at least one transition a day from above 0°C to below 0°C , Fig. 1) as a proxy for unpredictability (Table 1). This predicts the following rank order in dormancy: North > Central > East = Mountain = South.

Plant material

Mature seeds of *K. arvensis* were collected from five different geographical regions in Norway during the summer of 1998 (Fig. 1, Table 1). The seeds were combined into bulk samples, with each population containing seeds from one to four sites and from more than 50 individuals. For the field regeneration experiment, batches of 100 seeds from the Mountain population were sown within two weeks of collection (see below). The remaining seeds were stored in paper bags at 4°C until batches of 50 apparently ripe and undamaged seeds were placed in 100-mm petri dishes on moist filter paper for the phytotron experiments.

Fig. 1. The geographical distribution of *Knautia arvensis* (shaded) in Norway (Mossberg et al. 1992; Lid & Lid 1994). Locations of the populations used for the phytotron germination experiment (●), germinability after cold storage experiment (○), field regeneration experiment (■), and field regeneration observational study (□). The inserted panels summarise trends in diurnal temperature extremes through the year (data from the period 1960-1990; www.DNMI.no). Mean daily maximum (+1 SD) and minimum (-1 SD) temperatures are represented as solid (dotted) lines. Dashed horizontal lines are inserted to mark 0°C .



Phytotron germination experiments

Seed germination requirements and dormancy levels for four populations (Fig. 1) were tested in a series of phytotron experiments investigating the effect of (a) light and temperature; (b) fluctuating temperatures; (c) cold stratification and (d) dormancy breaking by means of gibberellic acid. Gibberellins serve vital physiological functions in seed germination (Vleeshouwers et al. 1995; Toyomasu et al. 1998) and may initiate germination in dormant seeds, or under suboptimal conditions (e.g. Bell et al. 1995, 1999). This treatment was included as an indication of whether any important environmental cues that break dormancy and initiate germination in *K. arvensis* were missed in our protocol.

In the temperature and light experiments (a, b) four constant temperatures (10°, 15°, 20° and 25°C) and a diurnal cycle (25° for 16h and 10°C for 8h) were compared in light and darkness. The 24h temperature sums of the 20°C and 25/10°C treatments are identical; by testing germination responses to fluctuating temperature against germination at 20°C we investigated the effect of diurnal variation *per se*. In the cold stratification experiment (c) unstratified seeds were set to germinate at 20°C in light and darkness, using germination of stratified seeds at 20°C as control. In the gibberellic acid experiment (d), the petri dishes were watered with 0.8% gibberellic acid, GA₃ and set to germinate in light at 20°C, using germination of stratified seeds at 20°C and

Table 2. Repeated measures ANOVA on the germination of *Knautia arvensis* from four populations (Mountain, North, East and Central) in Norway. The effects of (a) temperature and light; (b) diurnally fluctuating temperatures; (c) cold stratification and (d) dormancy breaking by means of gibberellic acid were tested. For experiments (b) and (c) controls were germinated at 20°C in light and darkness, for experiment (d) controls were germinated at 20°C and light. Significant treatment and interactions between treatments effects show the overall effects of treatments on the germination of *K. arvensis*, treatment × time effects signify an influence on the timing of germination, population × time effects signify differences between populations and population × treatment and population × time × treatment signify differential responses of populations to treatments. The differences between populations and the effects of time do not merit interpretation, as the former may be influenced by a number of factors including differences in viability between the seed batches and the latter simply reflects that germination accumulates through time (shown in italics).

Experiment	Source of variation	Model df	Error df	F-ratio	p
(a) Temperature and light	Temperature	3	41.5	82.1	< 0.0001
	Light	1	41.5	6.8	0.0128
	Temperature × light	3	41.5	1.1	0.3425
	<i>Time</i>	6	175	128.4	< 0.0001
	Time × temperature	18	175	13.6	< 0.0001
	Time × light	6	175	8.3	< 0.0001
	Time × temperature × light	18	175	2.0	0.0149
	<i>Population</i>	3	41.5	18.0	< 0.0001
	Population × temperature	9	41.5	5.7	< 0.0001
	Population × light	3	41.5	0.7	0.5426
	Population × temperature × light	9	41.5	0.4	0.9161
	<i>Population × time</i>	18	175	4.2	< 0.0001
	Population × time × temperature	54	175	1.4	0.0430
	Population × time × light	18	175	1.2	0.2313
	Population × time × temperature × light	54	175	0.4	0.9999
(b) Fluctuating temperature	Fluctuation	1	24	0.1	0.8345
	<i>Time</i>	6	144	190.3	< 0.0001
	Time × fluctuation	6	144	0.8	0.5614
	<i>Population</i>	3	24	22.1	< 0.0001
	Population × fluctuation	3	24	0.2	0.8845
	<i>Population × time</i>	18	144	6.8	< 0.0001
	Population × time × fluctuation	18	144	0.6	0.8671
(c) Cold-stratification	Stratification	1	32.7	66.6	< 0.0001
	<i>Time</i>	6	134	73.4	< 0.0001
	Time × stratification	6	134	12.1	< 0.0001
	<i>Population</i>	3	32.7	14.7	< 0.0001
	Population × stratification	3	32.7	2.8	0.0534
	<i>Population × time</i>	18	134	3.5	< 0.0001
	Population × time × stratification	18	134	1.1	0.3762
(d) Dormancy breaking (GA ₃)	Gibberellic acid	1	21.8	35.7	< 0.0001
	<i>Time</i>	6	86.1	58.1	< 0.0001
	Time × gibberellic acid	6	86.1	8.9	< 0.0001
	<i>Population</i>	3	21.8	10.5	0.0002
	Population × gibberellic acid	3	21.8	2.9	0.0609
	<i>Population × time</i>	18	86.1	2.3	0.0062
	Population × time × gibberellic acid	18	86.1	2.1	0.0139

light as the control. The factorial combinations of treatments were replicated two (a, b, c) or four (d) times for each population, for a total of 112 petri dishes and 5600 seeds. Germination was recorded and seedlings removed after 2, 4, 6, 10, 16, 24 and 32 days. For all except the immediate germination treatment (c), moist seeds were stratified in darkness at 4°C for two months prior to the experiments. Temperatures were regulated in specially constructed growth chambers. Light treatments were standard artificial greenhouse light for a photoperiod of 16h per day or darkness. For darkness, petri dishes were wrapped individually in two layers of aluminium foil and seeds were counted under a safe green light (< 0.05µmol m⁻² s). The experiments were carried out at the Centre for Plant Research in Controlled Climate at the Agricultural University of Norway. Repeated measures ANOVA on arcsine transformed data was used to test for differences in germination responses among treatments and populations (Sokal & Rohlf 1995). Days since onset and all experimental treatments were treated as class variables, and a first order autoregressive covariance structure assumed. All models were fitted using the procedure Proc mixed in SAS version 8.0 (Anon. 1999a).

Germinability after cold storage experiment

Seed germination after 8, 12, 16, 20, 24, 28 and 32 mo of storage at 4°C in darkness was tested for four populations (Fig. 1) in a phytotron experiment. Storage times were replicated twice, for a total of 56 petri dishes and 2800 seeds. Germinated seeds were counted and removed weekly for six weeks. The experiment was carried out at the Phytotron of the Department of Biology at the University of Tromsø, under constant artificial greenhouse light and 21°C. Because of problems with moisture regulation, the 12 and 16 month data were not used. To assess loss of germinability through time, a continuous exponential survival function was fitted to the data: $N = N_0 e^{-bt}$, where N is the number of seeds germinating at time t , N_0 is the number of seeds germinating at the first trial, t is time in months between N_0 and N and b is the seed loss rate (Cousens & Mortimer 1995). For comparative purposes the results were expressed as 'half-lives', which is the time – in months – until germinability was reduced by 50% relative to the first experimental date: $t_{1/2} = (\log_e 2)/b$. Among-population differences in loss of germinability over time were evaluated by one-way ANOVA on arcsine transformed germination data from the different storage times. Analyses were performed with SPSS 10.0 (Anon. 1999b).

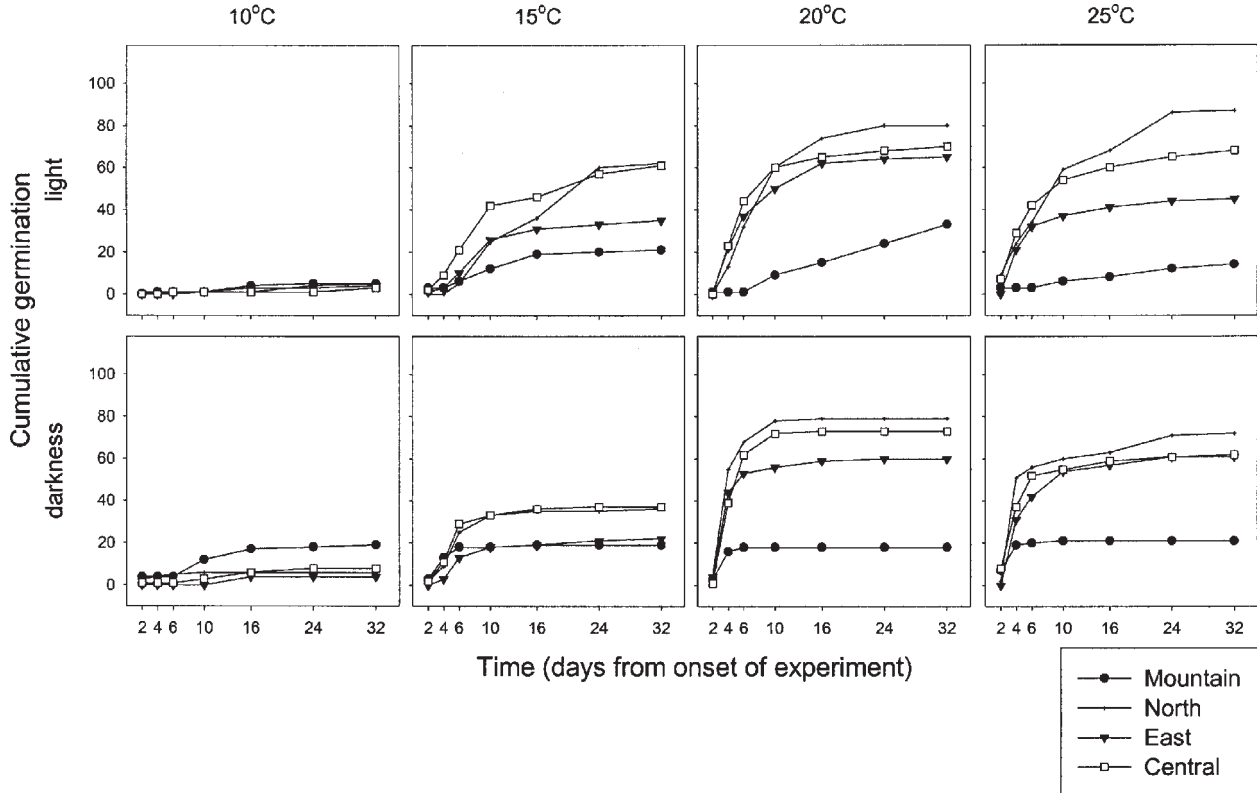


Fig. 2. Cumulative germination percentages for *Knautia arvensis* seeds collected from each of four populations under each of the eight light × temperature combinations in the phytotron.

Seed regeneration in perennial grassland

Data on the seed regeneration of *K. arvensis* in perennial grassland were obtained for two populations. In the North population, natural seed regeneration in the field was recorded in four 9m² permanent plots in September 1998, June 1999 and September 1999. At the Mountain population, a field regeneration experiment was initiated in October 1998 when batches of 100 seeds from this population were sown into 20 0.04 m² plots in a randomised block design with four treatments (turf stripped, vegetation cut at ground level, vegetation cut 5 cm above ground and standing vegetation) in five replicate blocks. The locations of individual seedlings were mapped early and late in the growing season for two years. In the experiment, treatment effects were tested for in mixed model ANOVAs with log_e (*n* + 1) transformed counts of new seedlings and survivors per plot per season as dependent variables, treatments as fixed factors, and experimental blocks as random factors (Proc mixed, SAS 8.0, Anon. 1999a).

Results

Knautia arvensis germinated under all experimental conditions, in the phytotron as well as in the field, but with considerable variation between populations, treatments and times (Figs. 2, 3, and 4). Germination percentages ranged widely among the petri dishes in the phytotron with 2–40%, 0–94%, 2–74% and 0–84% of the seeds germinating from the Mountain, North, East and Central populations, respectively. This means that in all populations a fraction of the seeds remained ungerminated even under the ‘best’ phytotron conditions. As the viability of these ungerminated seeds was not tested, we cannot determine whether they were still dormant, if they had died during the experiments or if there had been differences in initial viability. Because of

this, differences among populations in absolute germination are not very meaningful and will not be considered further (Table 2). Instead, we focus on the relative differences among populations in their responses to the treatments. In the statistical model, these effects are tested by the interaction terms population × treatment and population × treatment × time. Following the two major aims of the study, we start by presenting the overall germination responses of *K. arvensis* and relate these to the field germination results. We then move to evaluate the differences in phytotron responses among populations.

The relationship between phytotron germination responses and field behaviour

A conspicuous feature of the phytotron germination responses was the strong temperature response (Fig. 2; Table 2a); germination was high and fast at 20°C, slow at 15°C and very few seeds germinated at 10°C ($F = 82.1, p < 0.0001$). The differences in temperature response became more prominent through time ($F = 13.6, p < 0.0001$). The light regime was of much less overall importance and although it exerted a significant impact ($F = 6.8, p = 0.0128$), germination in light was less than 4% higher than germination in darkness after 32 days, giving no convincing evidence for upholding *Prediction 1* for *K. arvensis* (Table 2a; Fig. 2). Diurnally fluctuating temperature (25/10°C) did not increase germination beyond the 20°C levels (Table 2b), indicating that *Prediction 2* does not apply. Cold stratification substantially increased both germination rate ($F = 12.1, p < 0.0001$) and overall germinability ($F = 66.6, p < 0.0001$; Fig. 3; Table 2c) and after 32 days germination was 1.8 times higher (48% vs 27%) in stratified than unstratified seeds. This demonstrates that the seeds were weakly dormant when shed and that cold stratification contributed to breaking this dormancy, supporting *Prediction 3*. The addition of gibberellic acid (GA₃) to unstratified

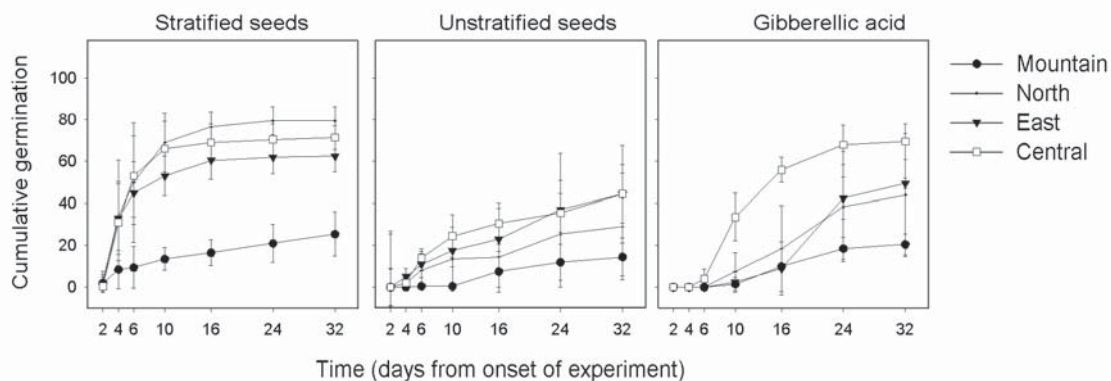


Fig. 3. Cumulative germination percentages at 20°C for *Knautia arvensis* seeds, collected from each of four populations, stratified (4°C for two months), germinated without any pre-treatment or watered with gibberellic acid.

seeds did not increase germination beyond the stratified seed levels (Fig. 3), suggesting that there are no additional unidentified germination requirements or sources of dormancy. The germinability of *K. arvensis* decreased substantially during cold storage (Fig. 4), but still 16.5% of the seeds germinated after 32 months and the estimated seed half-life for *K. arvensis*, based on three populations (North, East, and South), was 17.2 months. This supports *Prediction 4* about the potential for across year seed survival.

In the observational regeneration study at the North population, 1687 seedlings, corresponding to 16 m² per census, were recorded. In the field regeneration experiment in the Mountain population, 3.7% of the sown seeds were recorded as seedlings (73 in total), corresponding to 19 m² per census. In agreement with the phytotron results, *Predictions 1* and *2* were not supported in the field experiment, as the experimental microsite treatments (bare ground, vegetation cut 0 cm and 5 cm above ground and control) did not affect germination ($p = 0.9767$) or seedling mortality ($p = 0.9304$). In both studies, new seedlings were found at all census times: 27%, 38% and 35% in the North; 30%, 27% and 43% in the Mountains, at the autumn, spring and second autumn censuses, respectively. *Prediction 3* is therefore not supported in the field, a result that is inconsistent with the phytotron response (see above). The emergence of new seedlings for three censuses over two years after sowing in the field experiment is in agreement with *Prediction 4* and consistent with phytotron results.

Variation in phytotron germination responses among populations

Within the overall phytotron germination responses of *K. arvensis* described above, populations responded differently to some of the treatments. There were considerable differences in relative temperature responses among populations ($F = 5.7$, $p < 0.0001$). This effect became more pronounced with time ($F = 1.4$, $p = 0.0430$; Table 2a). The North, East and Central populations had relatively high temperature thresholds for germination relative to the Mountain population; i.e. they germinated poorly at 10°C, both in absolute numbers (4-6% vs 12%) and relative to germination at 20°C (12 to 16-fold vs 2-fold increase; Fig. 2). Fluctuating temperature (Table 2b) or light (Table 2a) responses did not, however, vary. Although marginally insignificant, there were also some differences in cold stratification responses ($F = 2.8$, $p = 0.0534$; Table 2c). The North population had the strongest relative response, a 2.7-fold (29-80%) increase in germination, whereas the effect was weaker, but still positive, in the Mountain, Central and East

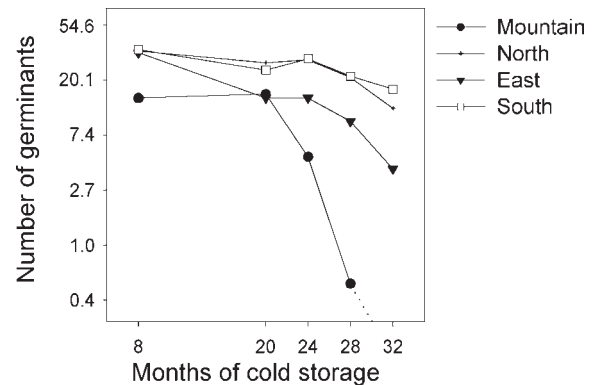


Fig. 4. Seed germination of *K. arvensis* from four populations (Mountain, North, East and South) after 8-32 months of storage at 4°C and darkness. The mean numbers of seeds germinated (exponential scale) in two replicate dishes are shown. No seeds germinated in the Mountain population at 32 months (curve indicated by the dotted line).

populations with 1.8-fold (15-26%), 1.6-fold (45-72%) and 1.4-fold (45-63%) increases, respectively (Fig. 3).

There was a trend of increasing difference in germinability among populations with the duration of cold storage (Fig. 4). The half-life estimates were 19.6 months for the North, 18.7 for the South and 15.1 months for the East population. The North, South and East populations could not be distinguished from each other initially (one-way ANOVA $p = 0.89$ and 0.58 at 8 and 20 months of storage, respectively), but differences emerged and grew stronger with time ($p = 0.07$, 0.14 and 0.003 at 24, 28 and 32 months, respectively). Half-life could not be estimated for the Mountain population because the data did not meet the assumptions of a negative exponential response through time. Germinability did seem to decline most dramatically in this population (Fig. 4).

Discussion

Can phytotron germination responses predict field behaviour in *Knautia arvensis*?

The phytotron experiments suggest that *K. arvensis* has a wide germination niche; the seeds germinated both with and without cold stratification, under a wide range of constant and fluctuating temperatures and in both light and darkness. The two field studies support this overall impression, seedlings are common within natural populations, appear in different microsites and throughout the growing season.

Predictions 1 and *2* state that germination requirements for light or diurnally fluctuating temperatures

will function as mechanisms for detecting ephemeral bare ground gaps in grassland vegetation (Rice 1985; Olff et al. 1994; Kotorová & Lepš 1999). Such responses were not found in the phytotron experiments, suggesting that these mechanisms are absent in *K. arvensis* (Figs. 2 and 3; Table 2). This was confirmed in the field, where germination was observed throughout the grasslands studied, with no detrimental effect due to the presence, absence or height of the grassland sward.

Furthermore, the subsequent survival of individual seedlings was also unaffected by sward characteristics, giving further strength to the interpretation that *K. arvensis* seedlings survive by tolerating, rather than avoiding, grassland sward competition. This agrees with Eriksson's (1989) prediction that clonals with 'repeated' germination should have high seedling competitiveness.

The results relating to *Prediction 3* were less straightforward. Although *K. arvensis* had a strong cold stratification response in the phytotron (Fig. 3; Table 2c), the field regeneration observational study and experiment gave no evidence for seasonally synchronised recruitment patterns. Although it is possible that the low temporal resolution of the field data masks a seasonal pattern, this is unlikely, given that similar numbers of new seedlings (with cotyledons present) were observed both in the late spring and in the early autumn at each of the two field sites. A lack of clear relationships between dormancy and germination timing in the field has previously been reported from temperate climates (Washitani & Masuda 1990; Schütz & Milberg 1997; Milberg et al. 2001). Schütz & Milberg (1997) suggest that a different germination syndrome, an inability to germinate at low temperatures, could contribute to the regulation of germination timing here. This, they argue, is because high temperatures would be unlikely to persist for sufficient time intervals to initiate germination during the unfavourable season. Such responses, often referred to as high temperature thresholds, were a conspicuous feature of our phytotron germination results (Fig. 2; Table 2a) and have previously been reported for northern, compared to southern, species in Britain (Grime et al. 1981) and for mountain compared to lowland populations in the Andes (Cavieres & Arroyo 2001).

Regarding the other predicted effect of dormancy, to facilitate seed carry-over across seasons, *Prediction 4*, our results were more consistent. In three independent experiments we showed that the seeds were partly dormant when shed (Fig. 3), that they could remain viable for extended periods (Fig. 4), and that carry-over actually occurred in the field as relatively high seedling recruitment was recorded both in the spring and autumn of the second year in the two-year field experiment (see also Willems & Bik 1998). Based on these results, a

working hypothesis for further studies is therefore that dormancy functions to delay germination and facilitate seed carry over in *K. arvensis*, rather than as a mechanism involved in the seasonal germination timing.

Are there differences in phytotron germination responses among populations, and if so, how do these relate to climate?

Data on variation in germination responses along climate gradients have accumulated over recent decades (e.g. Billings & Mooney 1968; Thompson 1975; Meyer et al. 1989, 1995; Meyer & Monsen 1991; Schütz & Milberg 1997; Andersson & Milberg 1998; Cavieres & Arroyo 2000; Navarro & Guitián 2003). The conclusions of these studies have been partly contradictory, leading some researchers to question the ecological interpretability of the among-population patterns (e.g. Schütz & Milberg 1997). We argue that these contrasting views can actually be categorized into the two alternative hypotheses outlined in the Introduction. Our phytotron experiments demonstrated considerable among population variation in germination responses (Table 2; Fig. 4) and several trends that relate to these hypotheses. Firstly, cold stratification responses are direct estimates of the dormancy levels in fresh seeds. The North population had a cold stratification response more than twice as strong as the other populations (Fig. 3), giving the relative rank order in dormancy North > Mountain = East = Central. Secondly, differences in the potential for long-term seed survival will have strong effects on the actual seed carry-over across years and hence on realised dormancy fractions. The North and South populations had a slower loss of germinability in cold stored seeds than the East and Mountain populations (Fig. 4) and if these results are taken into account then the relative rank order in dormancy becomes North > South > East = Central > Mountain. It should be noted that the relative rank position of the South population is only based on its performance in this experiment. Thirdly, high temperature thresholds for germination are a mechanism that may contribute to the regulation of germination during the cold season as 'functional equivalents' of dormancy (Schütz & Milberg 1997; Cavieres & Arroyo 2001; see discussion of *prediction 3*). The North, East and Central populations had higher temperature thresholds than the Mountain population (Fig. 2), so taking this response into account did not change the rank order of populations.

Overall, these dormancy-related responses were strong in the North, intermediate in the East, Central and South and weak in the Mountain population. This is almost the reverse of the order predicted by *Hypothesis 1*, and the hypothesis that dormancy increases with the

severity of winter climate is, therefore, not supported for *K. arvensis* in Norway. The results are more consistent with *Hypothesis 2* as the relative rank order of all populations, except the South, is consistent with the predictions based on the hypothesis that dormancy increases with the unpredictability of the winter climate. Clearly, more studies, involving a larger number of populations sampled along non-covarying gradients in climatic severity and predictability, are needed. Interestingly, *Hypothesis 2* and our results also conform to a prediction based on 'bet-hedging' theory, which states that dormancy fractions should increase as the environment becomes less predictable (Venable & Brown 1988; Philippi 1993a).

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